

Department of Science and Agriculture

BARBADOS.

**AGRICULTURAL
JOURNAL**

Vol. 5, No. 1.

January, 1936.

ADVOCATE CO., LTD.,—Printers & Bookbinders to the Government of Barbados.

DEPARTMENT OF SCIENCE AND AGRICULTURE

STAFF.

Director of Science and Agriculture .	R. W. R. MILLER, B.A., (Camb.) A.I.C., Dip. Agric. (Cantab.)
Assistant Director	Vacant.
Geneticist	A. E. S. McINTOSH, B.Sc., Ph.D., (Edin.)
Agricultural Chemist	S. J. SAINT, M.Sc., Ph.D., F.I.C. (London).
Assistant to Professor of Chemistry	T. A. E. BARKER, B.Sc., (London).
Lecturer in Natural and Agricultural Sciences	W. A. McD. PATERSON, B.Sc.
Entomologist	R. W. E. TUCKER, M.A., (Cantab.) B.Ed., (Capetown).
Veterinary Officer	Vacant.
Chief Analytical Assistant	C. A. COPPIN, B.A.Sc. (Toronto) A.I.C.
Plant Diseases Inspector	D. R. D. WILES, D.I.C.T.A.
Chief Clerk	H. O. RAMSEY.
Second Clerk	G. D. RAMSEY.
Third Clerk	C. G. HUMPHREY.
Fourth Clerk	A. S. INNISS.
First Field Assistant	C. B. FOSTER.
Second Field Assistant	H. E. WARD.
Third Field Assistant	C. C. CORBIN.
Fourth Field Assistant	F. E. BYNOE.
Analytical Assistant	H. A. JONES.
„ „	C. ST. A. ALLEYNE.
„ „	L. A. HALL.
Botanical Assistant	L. H. ATKINSON.
Foreman in charge of Nurseries ..	S. A. EASTMOND.

BRITISH WEST INDIES SUGAR CANE BREEDING STATION.

ADVISORY COMMITTEE.

Chairman	R. W. R. MILLER, B.A., (Camb.), A.I.C., Dip. Agric. (Cantab.), Director of Science and Agriculture, Barbados.
Members	SIR GEOFFREY EVANS, Kt., M.A., C.I.E., Principal of the Imperial College of Tropical Agriculture.
					HON. E. J. WORTLEY, O.B.E., Director of Agriculture, Trinidad.
					A. C. BARNES, B.Sc., F.I.C., A.M.I. Ch.E., Director of Agriculture, Jamaica.
					G. A. JONES, Dip. Agric. (Bangor), Commissioner of Agriculture of the Agricultural Advisory Department of the Imperial College of Tropical Agriculture.
Geneticist	A. E. S. McINTOSH, B.Sc., Ph.D., (Edin.).
Assistant Botanist	G. C. STEVENSON, B.A., A.I.C.T.A.
Field Assistant	J. M. CAVE, D.I.C.T.A.
Statistical Clerk	A. D. HERBERT, D.I.C.T.A.

BOARD OF AGRICULTURE.


The members of the Board of Agriculture, appointed for the Legislative Session, 1934—35, were as follows:—

The Honourable S. C. Thorne, M.L.C., Chairman.
The Honourable G. L. Pile, B.A., M.L.C.
Sir George Walton, Kt., M.C.P.
G. D. L. Pile, Esq., B.A., M.C.P.
G. C. Mahon, Esq., M.C.P.
Henry Alleyne, Esq., M.C.P.
H. A. Arthur, Esq., M.C.P.
E. S. Robinson, Esq.

The Director of Agriculture is an *ex officio* member of the Board.

TABLE OF CONTENTS

	Page.
Parasites introduced into Barbados for control of Insect Pests ..	1—22
Investigations into the use of the Zeiss Hand Refractometer ..	23—39



Digitized by the Internet Archive
in 2025

PARASITES INTRODUCED INTO BARBADOS FOR CONTROL OF INSECT PESTS.

BY

R. W. E. TUCKER, M.A., B.Ed.

The parasites and predators which have been introduced within recent years into Barbados are as follows:—

- I. For control of *Diatraea saccharalis* Fabr., the small moth borer of sugar cane:

Ipobracon grenadensis, Ashmead.

Microdus diatraea, Turner.

Lixophaga diatraea, Townsend.

Paratheresia claripalpis, Van der Wulp.

Trichogramma minutum, Riley. Biological races of this species have been introduced from Louisiana, California, Massachusetts, Mexico, Antigua, St. Lucia and Montserrat, together with an Antigua by Barbados cross supplied by Dr. S. C. Harland, and all have been tested for mass breeding purposes.

- II. For control of *Diaprepes abbreviatus*, the root borer of sugar cane:

Tetrastichus haitiensis, Gahan.

Ufens osborni, Dozier.

- III. For control of *Lachnosterna smithi*, Arrow, brown hardback pests of sugar cane:

Elis ephippium (*xanthonotus*), Rohwer.

Elis haemorrhoidalis, Fabr.

Campsomeris tricineta, F. (*pyrura*, Rohwer).

Campsomeris trifasciata, Fabr.

Pyrophorus luminosus, Illiger.

- IV. For the control of *Pectinophora gossypiella* Saund, pink bollworm of cotton:

Microbracon kirkpatricki, Wilk.

- V. For the control of lepidopterous defoliators of cotton and sweet potato:

Compsilura concinnata. Meigen.

I. For Control of *Diatraea saccharalis*, Fabr., the Small Moth Borer of Sugar Cane.

The control of *D. saccharalis* in cane is recognised as being a complicated and stubborn problem, and the factor of artificial or introduced biological control is only one of several factors which influence the prevalence of, and damage caused by, this pest.

Further, although *D. saccharalis* is widely spread throughout the North American Caribbean and South American sugar growing areas, it has different natural biological complexes in different areas, particularly in the presence, or absence, of various larval and pupal parasites.

It is, therefore, natural that efforts towards the control of *D. saccharalis* should be made by the transference of larval and pupal parasites from areas where such exist, to areas where none, or different kinds of parasites, exist; and as early as 1915, the Tachinid *Lixophaga diatraea* was obtained in Cuba for shipment to the sugar areas of Louisiana, and other efforts have followed this.

One of the pioneers in the work was H. E. Box, and, in the course of obtaining larval parasites in Venezuela and the Guianas for shipment to Porto Rico, he was able to supply Barbados with the Braconids *Ipobracon grenadensis* and *Microdus diatraea*.

Mr. Box visited Barbados in 1927 and handed over a supply of the above Braconids, which were artificially propagated in a large field cage (2) erected at the Codrington Experimental Station. Subsequently a consignment of around two thousand *Ipobracon* was sent from British Guiana, of which around 80 per cent. arrived alive in Barbados.

From this adequate material colonies were reared and distributed in the field, and, following Mr. Box's advice, large numbers of *Cordia interrupta* bushes were grown and distributed over the Island to act as food plants for these parasites.

When the writer arrived in Barbados in July 1928 to carry out biological measures of control for cane pests, the first investigation was that of ascertaining whether these parasites had become established, and whether they could be recovered in the field: also, to ascertain whether any other parasites of *Diatraea saccharalis* were present.

As the writer was informed that the introduced Braconids had been observed in large numbers around *Cordia* bushes late in 1927, it was with every expectation of finding them that search was made from August onwards in 1928 and early in 1929. Bored canes were dissected to find cocoons, and large quantities of bored cane, particularly from Codrington, were placed in cages specially constructed for the purpose, so that emergence of parasites could be observed. Plenty of *D. saccharalis* were obtained in this manner, but no Braconids or other larval or pupal parasites. Repeated observations were also made at *Cordia* bushes around cane fields at Codrington and elsewhere, and collections made of insects attracted to, or found feeding on, *Cordia* bushes: again, however, no introduced Braconids were found.

Since 1928, *Cordia* bushes have been kept under observation, and many thousands of canes have been dissected, but no recoveries of these introduced parasites have been made.

It is therefore concluded that the Braconids introduced in 1927 failed to become permanently established, though introduced under favourable circumstances.*

The next parasite to be introduced into Barbados for the control of *D. saccharalis* was the Tachinid *Lixophaga diatraea* from Cuba, which had already been introduced into Louisiana and into British Guiana, and in 1930 and 1932 into Antigua.

Puparia of this parasite were obtained in Cuba early in 1930 by Dr. J. G. Myers, and on May 3rd a consignment of 186 puparia was received in Barbados.

A large cage, 30 feet by 30 feet by 8 feet, was erected over young infested canes in an open canefield, and, of the total of 112 *Lixophaga* adults which emerged from the above puparia, 82 were placed in the cage, with additional cut infested shoots, and 30 were liberated some distance away in the same field.

At the end of March, 1933, a systematic search was made for *Lixophaga* from twenty-two fields within a two mile radius of the area of liberation. From a total of 2,640 dead hearts dissected, no *Lixophaga* were recovered, nor had other previous but more restricted searches produced any results.

It was concluded, however, that the above liberation was on too small a scale, and that better results could be expected from the artificial rearing of the parasite in the laboratory and the colonisation of definitely mated females which had been allowed to develop to nearly their full period of gestation in the laboratory.

This method had been fully carried out in Antigua by Box in 1932 and 1933, following the laboratory rearing technique elaborated by L. C. Scaramuzza in Cuba, and proof had been obtained that the parasite was established in the field.

Arrangements were therefore made in May, 1934, for the writer to visit Antigua for the purpose of obtaining a small colony of the acclimatised flies and to practise the laboratory rearing technique, so that from a small colony, adequate numbers could be reared and liberated in Barbados.

During the latter part of May and early part of June, a total of 78 *Lixophaga* puparia was obtained in Antigua from which 37 females and 35 males were bred.

From 28 mated females, 1,555 inoculations of *Diatraea* larvae were made in Antigua, and the resulting parasitised material was brought down to Barbados. A recovery of 735 *Lixophaga* puparia (47.2 per cent.) was obtained in Barbados from which adequate breeding material was assured, and a surplus left over for immediate liberation.

* When dissecting large numbers of dead hearts to obtain *Diatraea* larvae for breeding *Lixophaga* early in 1935, the Braconid *Microdus stigmaterus* (identified by Dr. J. G. Myers) was obtained. Altogether 7 *M. stigmaterus* were obtained out of a total of 38,132 *Diatraea* larvae and pupae, which is 0.018% parasitism. This, however, is most probably not an introduced parasite.

Some difficulty was experienced at first in obtaining adequate *Diatraea* larvae for laboratory inoculation with *Lixophaga* maggots in Barbados, partly because there was an abnormal preponderance of living *Diatraea* pupae over living larvae, and also because the larvae, when removed from their shoots, pupated shortly afterward and without feeding though often not fully grown.

These abnormal conditions, which appear to be attributable to the prolonged low rainfall in 1934, made the accumulation of stocks of *Diatraea* larvae a difficult matter.

Nevertheless, from the latter part of July to the beginning of October, 1934, a total of 3,229 fertile *Lixophaga* females, and 851 males was distributed mainly in the central hillier and wetter areas of the Island. A general recovery of 60 per cent. to 70 per cent. was obtained in the laboratory from inoculations of *Diatraea* larvae with *Lixophaga* grubs.

Although much less rain than usual fell in Barbados in 1934, the areas of liberation received sufficient light showers to keep the cane stalks and shoots moist at intervals during the above period of liberation, so that, as the female *Lixophaga* liberated were ready to deposit larvae soon after liberation, the above numbers should have secured some measure of establishment in the field.

As a precaution, however, a laboratory colony was maintained throughout 1934—35, so that large scale breeding of this parasite could be resumed in the spring of 1935. At this period *Diatraea* larvae are more easily obtained in sufficient quantities from young infested cane shoots, and laboratory rearing for liberation of mated females becomes a practicable proposition.

The areas of liberation were searched from time to time in 1934 for traces of activity on the part of the colonised *Lixophaga*. Two larvae were recovered from bored shoots, and reared to adult *Lixophaga*, and one puparium was recovered from Codrington at the end of November, 1934. Subsequently two more puparia were recovered from Codrington in December, 1934, and one living and two empty puparia from Waterford (near Codrington) in February, 1935. No other positive results were obtained in 1934.

In the early months of 1935, tens of thousands of dead hearts were dissected to obtain *Diatraea* larvae in which to mass rear the laboratory colony of *Lixophaga* maintained from 1934.

Altogether, 38,132 *Diatraea* larvae and pupae were obtained during this work, and careful watch was kept for *Lixophaga*. Eleven were obtained from this mass of field material.

The liberations of *Lixophaga* made in 1935 totalled 11,594 (6,793 females and 4,801 males), and the area of liberation was confined to the Applewhaites, Walkes Spring, Mount Wilton and Lammings area. The concentration of the above liberation in this area should certainly have sufficed for the establishment of this parasite. It is too early as yet to state definitely that this introduction and adequate colonisation has failed, but examinations carried out in November and December of 1935 in which discarded bored top plants and portions of cane were dissected in the fields where cutting for plants was taking place at Lammings and

Mount Wilton, and in the yards at Walkes Spring and Applewhaites, produced one larva only of *Lixophaga*, found at Applewhaites. No empty pupal cases of *Lixophaga* were found, nor were any recoveries obtained from nearly 100 *Diatraea* larvae, examined or brought back to the laboratory for pupation. The usual recoveries of empty *Diatraea* pupal cases, of living *Diatraea* pupae and *Diatraea* larvae killed by *Cordyceps* were obtained in these examinations, and in examinations of fields of standing canes in the area of liberation.

No strong hopes are entertained, therefore, that a field survey of young canes in 1936 will show a satisfactory establishment of *Lixophaga* in Barbados. This is regretted, because the economic establishment of *Lixophaga* in Barbados has always been a desired object, and no pains have been spared to make the introduction and establishment a success†.

If the above negative indications are reversed at any future date, and *Lixophaga* eventually proven to be an established and measurable agency of control over *Diatraea saccharalis* in Barbados, the fact will be given full acknowledgment.

Coincident with the introduction and laboratory rearing of *Lixophaga* in 1934, consignments of *Paratheresia claripalpis* puparia were received from Trinidad through the kind offices of Mr. F. W. Urich of the Imperial College of Tropical Agriculture.

This Tachinid is more prevalent in Peru and Argentine than in Trinidad, but supplies are more readily obtained from the latter Island.

In Peru and Argentine it is recorded as an effective larval parasite of *D. saccharalis* and considerable supplies have been obtained there in recent years by Mr. H. A. Jaynes for shipment to Louisiana: shipments have also been made from Trinidad to Cuba.

Paratheresia is considered as being possibly more suited and adaptable to dry areas than *Lixophaga*, and it was planned to make liberations in a limited section of the drier areas of Barbados and to watch its progress. Unfortunately, it was found to be more difficult to rear in the laboratory than *Lixophaga*, and either died within a few hours or days of emergence without mating, or, if mated, succumbed before the period of gestation was completed. From one mated female fly, which died just prior to the full gestation period, three weak living maggots were recovered by dissection, and from these one successful inoculation was made. Altogether, 195 *Paratheresia* puparia were received in four widely separated sendings; from these, a total of 123 flies (78 females and 45 males) was obtained; the maximum number hatching at one period being 4 females and 8 males.

The laboratory propagation was, therefore, a failure, and only 24 mated, and presumed mated, females and 4 males were liberated in the field, from which no recoveries have been made.

From a final consignment of *Paratheresia* puparia received from Trinidad towards the end of October, no *Paratheresia* at all hatched out, but from some puparia numerous hyperparasites hatched.*

† In March 1936 a preliminary survey of the heavily colonised Applewhaites to Lamings Valley has given inadequate and disappointing recoveries of *Lixophaga* from young cane fields.

* *Trichopria (Planopria)* sp. probably new. Identified I. I. E.

In view of the fact that Scaramuzza reports (by correspondence) success in the laboratory rearing of this parasite in Cuba, it is possible that the lack of success in Barbados was due to inexperience of the proper technique, and that the routine which gave excellent results with *Lixophaga* needs special adaptation for *Paratheresia*. It is hoped to make a further attempt with *Paratheresia* at the conclusion of the *Lixophaga* breeding campaign in 1935.

Turning now to *Trichogramma minutum*, it may seem strange to place this parasite in the list of those introduced into Barbados, seeing that it is indigenous not only to Barbados but to all cane growing areas in the West Indies.

Nevertheless, the work of Dr. S. C. Harland in Trinidad (4) and of S. Flanders in California (3) has shown that not only do different biological races of *T. minutum* exist, but that these races differ in adaptability and in effectiveness; and, through the courtesy of the above workers, several biological strains have been tried out in Barbados.

When it was decided in 1928 to use mass reared *T. minutum* as a control over *D. saccharalis* in Barbados, it was found that the first local race of *Trichogramma* tried out did not take readily to *Sitotroga* eggs as hosts, and it was some few weeks before a breeding stock could be built up from local material. Application was therefore made to Dr. H. Spencer who was at that period working on a similar project in Louisiana and he very kindly sent considerable consignments of the Louisiana race of *T. minutum*. These *Trichogramma* were found to breed readily on *Sitotroga* eggs, and to be effective also in parasitising *Diatraea* eggs from the field. The grey Louisiana strain was, therefore, at that time adopted as the mainstay for mass production of *Trichogramma*, but has since died out, and has been replaced by other strains as described.

Later on, through the courtesy of Mr. S. Flanders of the Riverside Experiment Station, California, and of Dr. S. C. Harland, Empire Cotton Breeding Station, Trinidad, the following races of *T. minutum* were introduced into Barbados and experimented with:—

Massachusetts yellow short cycle: California transition long cycle: California dark long cycle: California dark short cycle: Mexican dark short cycle: St. Lucia: and a parthenogenetic strain developed from St. Lucia by Dr. Harland: Montserrat race: and an Antigua by Barbados cross.

Of these introductions, the Massachusetts strain is still maintained and contributes a portion to liberations made in Barbados.

The California transition long cycle strain proved successful and forms a considerable portion of liberations made.

The California dark long cycle and dark short cycle were not successful; the Mexican strain did not hatch.

The St. Lucia ordinary strain failed to oviposit in *Sitotroga* eggs and died out, and the St. Lucia parthenogenetic strain, though amenable to laboratory rearing, gradually died out. The Montserrat race also failed to breed in *Sitotroga* eggs and was very sluggish in action.

The Antigua by Barbados cross produced by Dr. S. C. Harland was, however, successful, and is also still maintained in mass breeding work. The remaining and principal strain used in mass breeding work is the local Barbados race.

The mass breeding releases, therefore, are mainly local Barbados, California transition long cycle, Barbados by Antigua cross and Massachusetts yellow short cycle.

The principles underlying the artificial breeding and releasing of a natural parasite and the experimental and crop result data which have thereby been obtained have been discussed in other papers, and the efficiency of *Trichogramma* as a primary parasite has been demonstrated by the recent work of Dr. G. Salt (6).

II: The Control of *Diaprepes abbreviatus*.

There are no parasites of any stage of this serious pest of cane in Barbados, and therefore the opening for the introduction of any parasite from elsewhere appeared promising. When therefore it was shown through the work of Dr. G. N. Wolcott that an egg parasite, *Tetrastichus haitiensis*, Gahan, of *Prepodes quadrivittatus*, Olivier, existed in Haiti, it was planned to introduce this parasite into Barbados, against the allied *Diaprepes abbreviatus*.

As in the case of *Diatraea* parasites, nothing could be done at first, owing to the fact that the writer was not free to obtain parasites from other countries until an adequate entomological organisation existed for receiving, breeding and distributing them in Barbados.

The arrival of Dr. J. G. Myers in the West Indies, on a mission connected with the search for and distribution of beneficial insect parasites, solved this difficulty, and his discovery that *T. haitiensis* parasitised the eggs of *Diaprepes famelicus* in Montserrat increased the probability that the parasite would be effective in Barbados.

In May, 1931, Dr. Myers made arrangements to visit Haiti where more abundant supplies of *T. haitiensis* could be obtained than in Montserrat, and it was hoped to obtain supplies of this parasite in June, when *Diaprepes* is normally most abundant in Barbados, and when there is the maximum quantity of eggs laid in the crossed tips of the young and growing cane.

As so often happens, however, seasons of insect abundance do not coincide in different areas, and Dr. Myers wrote from Haiti at the end of June that he had the utmost difficulty in obtaining *Diaprepes* in Haiti on his arrival, and that during a few days *en route* at Puerto Rico, both he and Dr. Osborn found that *Diaprepes* had barely begun laying and parasites were unobtainable.

By the end of June, *Diaprepes* was more abundant in Haiti, and the first consignment of parasites was received in Barbados on July 19th, having been sent to Antigua by aeroplane from Haiti, and from Antigua to Barbados by steamship.

A second consignment arrived on July 26th, having reached Trinidad by air mail on July 21st.

A third consignment, also *via* Trinidad, arrived on August 9th and a fourth consignment on August 23rd, 1931.

In all cases the material received was field collected, and, in the case of the three consignments, received *via* Trinidad, a considerable proportion of the parasites had already hatched out. The latter received immediate attention in the shape of food and water, and records were made of their subsequent actions, and of all emergences from the above material.

From the first consignment, a total of 39 parasites hatched; from the second a total of 60 was obtained, of which, as stated, a fair number had hatched on arrival. None of this latter consignment appeared at all vigorous, and by August 5th, ten days after arrival, the majority had died. From the third consignment a total of 134 parasites was obtained, of which 65 had hatched prior to arrival, and from the fourth consignment 66 were obtained of which 50 had hatched *en route*.

Dr. Myers had previously supplied the information that *T. haitiensis* had been found amenable to laboratory propagation, and that it would parasitise the eggs of *Diaprepes* laid between slips of glazed paper or waxed paper. At the same time it was stated that Dr. Osborn had found a giant Trichogrammatid parasitic on eggs of *D. abbreviatus* in Puerto Rico, which was also amenable to laboratory rearing, and that Dr. Myers would re-visit Puerto Rico and endeavour to obtain this parasite for Barbados.

Work was therefore started on laboratory rearing the *T. haitiensis* material from Haiti.

Parasites known to be fed and mated were supplied with *Diaprepes* eggs laid by caged beetles, between slips of waxed paper, and also with *Diaprepes* eggs laid between cane tips in the field and in the laboratory. Exposure to direct sunlight, diffused sunlight and shade made no appreciable difference to the behaviour of the parasites. Attempts at parasitising the eggs supplied were carefully watched, and it was found that difficulty was experienced by the parasite in inserting the ovipositor in the glazed surface of the waxed paper: subsequently, a softer matt surface paper was used with much greater success. The eggs laid between cane tips were never parasitised, though female *T. haitiensis* were frequently seen to wander over and explore the cane leaf surface.

Examination of the records kept shows that on August 17th twenty-six days after breeding operations started, a total of sixteen laboratory reared parasites only had been recovered, and these are recorded as being smaller and slenderer than the original stock received.

By August 26th a total of 110 laboratory reared parasites had been obtained after 35 days of breeding work, and the use of 268 parasites from Haiti, and about 18,000 *Diaprepes* eggs. Of the above laboratory total, 50 only were on hand for stock at that date, the remainder having been liberated at Lancaster Plantation, St. James, and Boarded Hall Plantation, St. George.

By September 7th the laboratory total of all generations had reached 185, of which only 12 were surviving and these were liberated in the field at Lancaster.

The third laboratory generation started to emerge on September 8th, but recoveries were so poor that to prevent extinction, all available parasites were put out in the field.

On August 23rd a fourth and final consignment of field material was received via Trinidad from Dr. Myers in Haiti, over 50 having hatched prior to arrival.

As the above records show, it was evident by then that the material from Haiti did not increase in a sufficiently successful manner to warrant continued laboratory breeding, so, after feeding and mating the above consignment, the parasites, to the total of 66, were liberated at Lancaster and Boarded Hall in the same areas as previous liberations.

Breeding records also show that the life cycle of the Haitian material in the laboratory was very erratic; the usual period was around 18 days, but some emerged after 14 days, and some took as long as 28 days, and in most cases the progeny appeared smaller and weaker than the stock received.

The next phase in the introduction of *T. haitiensis* started with the receipt on August 28th, 1931, of a consignment of field material from Puerto Rico, with cabled instructions from Dr. Myers to retain and breed only the giant Trichogrammatid (*Ufens osborni*) discovered by Dr. Osborn.

On opening up the consignment, between two and three hundred *Tetrastichus* only were found hatched out, which, according to instructions, were destroyed. A cable was however sent asking in what proportion the Trichogrammatid could be expected, what were relative life cycle periods, and asking for field, or other, data. A cabled reply from Dr. Myers stated that the information was not available as Osborn's work was insufficient: which in view of the recent discovery of this parasite is understandable. The material was, therefore, closely watched for emergences of the Trichogrammatid. On August 29th and 30th *Tetrastichus* only emerged and were destroyed. On August 31st three *Ufens osborni* emerged, were removed to a separate tube, fed, watered and supplied with *Diaprepes* eggs. Of the *Tetrastichus* emerging on that date, half were destroyed, and the remainder (about 100) were retained for comparative laboratory breeding tests.

On September 1st, only one *Ufens osborni* had survived out of the three mentioned above, and five were found hatched, but dead, in another tube of Puerto Rican material.

On September 3rd one more *Ufens osborni* hatched out: this one survived about 24 hours.

By September 30th there had been no further hatchings of *Ufens* and no parasitism of eggs by the two short-lived survivors noted above. The introduction of *Ufens osborni* which has since been shown to be largely, if not entirely, a secondary parasite, was therefore unsuccessful.

In the meantime, the *Tetrastichus* from Puerto Rico were breeding rapidly and successfully in the laboratory, under exactly the same routine as applied to

the Haitian material. This routine consisted in transferring newly hatched parasites to 6-inch by 1-inch glass tubes with gauze caps, supplying with water on the caps, and providing split raisins in the tubes for food. Under these conditions, the parasites mated readily. *Diaprepes* eggs laid between soft matt paper were obtained fresh daily from caged beetles (cold storage of *Diaprepes* eggs was found useless), and the egg masses clipped out of the paper and gummed on sheets of thin cardboard two of which fitted exactly in the tubes.

These eggs were readily attacked by *Tetrastichus*, and were removed daily, the sheets dated, and fresh material supplied.

An excellent recovery of parasites was invariably secured, and the life cycle was steady around 16 days. By the time the 3rd generation was in full swing, 1,000 parasites a day were being obtained and by October 26th, 19,000 had been liberated in the field. Altogether, 22,000 were liberated in 1931, and about 11,000 in July 1932 from fresh supplies obtained from Puerto Rico through the courtesy of Dr. M. Leonard and Dr. G. Wolcott. Further liberations were made in 1933 from material sent down from Puerto Rico by the writer.

The vigour of these parasites from Puerto Rico, and their apparent greater size and different laboratory reaction compared with the Haitian material, appeared to indicate possible different species, but subsequent identification proved them to be the same *T. haitiensis*. Before making field liberations, enquiries were made of Dr. Myers as to any possible objections to liberating the Puerto Rico *Tetrastichus*; there were, however, no objections.

Attempts were made from time to time to recover this parasite in the field, and thousands of egg masses were examined. As no recoveries were made, and as all attempts to get the parasite to breed in field collected egg masses laid in cane tips had failed, it was decided to test *T. haitiensis* more definitely with *Diaprepes* eggs in cane. Canes in pots, with young shoots of a size to go into a large laboratory cage, were first exposed to *Diaprepes* beetles and egg masses obtained between young cane leaves.

The beetles were then removed from the cage, and a number of mated *Tetrastichus* placed in the cage. The parasites were seen to wander over the cane leaf surface above egg masses, apparently endeavouring to oviposit in the egg masses. Eventually, a recovery of 14 parasites from the material was obtained, which was, however, a poor recovery, and showed that, despite the eggs being laid between fresh young cane leaf tips, *T. haitiensis* parasitised very few, despite every opportunity to do so.

Other attempts with newly gathered field material, kept fresh in jars of water, and with *Tetrastichus* confined in tubes over the egg masses, failed to give results except in one instance in which the cane tips had been pulled slightly apart and eggs exposed.

There appears therefore, to be a possibility of parasitism of *Diaprepes* eggs newly laid in young and tender cane tips, but very little possibility of parasitism in older, tougher leaves, which may explain the absence of field recoveries, despite the numbers liberated.

Evidence which appears to confirm this was obtained by the writer during a month's visit to Puerto Rico in June and July, 1933.

Diaprepes was found in abundance, and plentiful parasitised material was obtained from young citrus orchards, but in no case was parasitised material found in cane tips, though *Diaprepes* egg masses were obtained from canes growing alongside a heavily infested and parasitised young citrus orchard at Florida (Puerto Rico), whilst in an old citrus orchard near San Juan, in which canes were inter-planted with the citrus, *Diaprepes* eggs were obtained on the citrus, but not from the cane. At Aguirre and Guanica, in the South of Puerto Rico, *Diaprepes* egg masses were obtained from canefields as in Barbados, but in no case was *Tetrastichus* or any other parasite recovered, from these egg masses.

The searches for egg masses in canefields in Puerto Rico were admittedly not extensive, but whereas an abundance of *T. haitiensis* in citrus was obtained and sent to Barbados where it was bred and liberated in 1933, and small numbers of *Ufens* and a species of *Horismenus* also found in *Diaprepes* eggs in citrus, no parasitism was found in *Diaprepes* eggs in cane in Puerto Rico. In conjunction with field and laboratory experience in Barbados, it is therefore concluded that *T. haitiensis* does not or cannot, normally attack *Diaprepes* eggs when laid in cane.

The results therefore of the introduction of *T. haitiensis* are that, though considerable numbers were bred and liberated in 1931, 1932 and 1933, no field recoveries have yet been made despite frequent searches.

The introduction of *U. osborni* was a failure from the start, and was not repeated, especially as so little was then known of its status and effectiveness as a primary parasite of the eggs of *D. abbreviatus*.

III. The Control of *Lachnosterna Smithi*, the Brown Hardback Pest of Sugar Cane in Barbados.

This pest is fairly widely spread in Barbados, and the damage caused by it is erratic: that is to say, isolated fields or groups of fields in various districts are sufficiently badly attacked to cause noticeable and serious damage; and, in other cases, the pest, though present, does not cause serious or noticeable damage, though most probably influencing the ultimate yield of the fields. Ratoon fields are usually most heavily attacked, and in cases where stunting of canes and slight yellowing of growth is noticed, fifty or more *Lachnosterna* grubs have been obtained per cane hole.

There exists in Barbados a Scoliid parasite of the grubs or larvae of *Lachnosterna smithi*, namely, *Tiphia parallela*, Sm., but the writer has been able to obtain no evidence in the field that it is either abundant all over the Island, or effective in the extent of its parasitism. Considerable collections of *Lachnosterna* grubs have been made in the field from time to time, but in no case has an extensive or economically effective parasitism by *Tiphia* been found.

The highest parasitism found in the course of digging in several fields and obtaining considerable quantities of *Lachnosterna* grubs was eight per cent.: the usual sum of parasitism is 0 per cent.: with an occasional field of 2 per cent. up to 5 per cent. parasitism. Under these circumstances, although occasional fields can doubtless be found in which prolonged search would yield some hundreds of *Tiphia* cocoons and parasitised larvae it is not considered that *T. parallela* is an effective check on *L. smithi* in Barbados.

When, therefore, an opportunity occurred of collaborating with Mr. W. F. Jepson from the Farnham Royal Laboratory of the Imperial Institute of Entomology, who was searching in Puerto Rico for parasites of *L. smithi* on behalf of Mauritius, an attempt was made to secure beneficial parasites for Barbados.

The writer, therefore, proceeded to Puerto Rico in June, 1933, for the purpose of working with Mr. Jepson on parasites of the brown hardback, and at the same time to procure further supplies of *T. haitiensis* for Barbados, and to gain some knowledge of its local habits and status, and so to examine, as far as possible, the situation with regard to *Lixophaga* in Puerto Rico.

As a result of extensive searches for Scoliids, shipments of the following parasites were made by air mail and steamer to Barbados:—*Elis ephippium*, *Elis haemorrhoidalis*, *Campsomeris tricineta* and *Campsomeris trifasciata*.

Of these, *E. haemorrhoidalis* was very scarce during the writer's period in Puerto Rico, namely, from mid-June to mid-July, but, subsequent to the writer's return to Barbados, Mr. Jepson was able to send an adequate shipment down by air mail of which those which survived were kept for laboratory rearing.

One shipment of Scoliids collected by Mr. Jepson and the writer was made from Puerto Rico to Barbados, and one was brought down by the writer: Mr. Jepson also sent a consignment, mainly *ephippium*, in addition to the *haemorrhoidalis* mentioned above. In all cases the shipments were made by air mail to Trinidad and by steamer thence to Barbados.

The containers consisted of paraffin tins, into the open top of which a wooden frame three-quarters to an inch wide and half an inch thick was fitted. On to this a square of wire mosquito gauze was placed carrying a square of butter muslin and strips of wood corresponding with the above frame nailed over this to keep both in place. A cross piece of wood, complete with handle, finished the container. Inside, and fastened by wire passed through holes punched in the sides of the tin, were a bottle containing water and a wick to supply adequate moisture, and a piece of hardwood bored with thimble-like holes, into which were packed a mixture of sugar and honey for food. Damp moss, kept in place by wire gauze, was placed at the bottom of the tins, and some food plant, *Mitracarpus portoricensis*, and festoons of paper were placed in the tins to provide footholds for the wasps. Containers such as these accommodated numbers of wasps, and the mortality in transit was not great.

The first consignment, sent towards the end of June, 1933, consisted of 50 *Elis ephippium* females, 15 *Campsomeris trifasciata*, and five *Campsomeris tricineta*, together with sufficient males of each species, making over 100 in all. This arrived in Barbados on July 5th, with 19 *ephippium* dead (males and females) and 9 *trifasciata* and *tricineta* dead; that is, a total of 28 out of 100.

Of *ephippium*, 16 were kept for laboratory breeding and 34 were liberated at Mangrove Plantation; of the other species, 4 were kept for laboratory breeding and 15 were liberated at Stepney Plantation.

The preliminary breeding work was carried out on the plan for Scoliid breeding as given by E. Jarvis in "Tropical Agriculture" (5).

On the writer's return from Puerto Rico on July 19th, a further supply of 152 Scoliids was brought to Barbados via Trinidad: namely, 130 *E. ephippium*, 14 *trifasciata* and 8 *tricincta*; of this, a total of 34 died *en route*. Twenty *ephippium* females and a few males were kept for breeding purposes, as well as 5 each of *tricincta* and *trifasciata*, and the remaining Scoliids were liberated on the plantation at the Government Industrial School in an area from which good supplies of brown hardback grubs were being obtained.

Subsequently, Mr. Jepson sent a consignment of 28 females and 5 or 6 males of *E. haemorrhoidalis* which he had specially collected in order to compensate for the deficiency in this species at the period when the writer's collections were made. Of the consignment, only 13 arrived alive, though the packing was in every way normal. These were used for laboratory breeding.

Towards the end of August, Mr. Jepson sent a further consignment of 75 Scoliids, mainly *E. ephippium*, of which 25 *ephippium* were kept for laboratory breeding, and the remainder liberated at Dodds and Summervale.

The first duty to be undertaken by the writer on his return from Puerto Rico, was to find and obtain adequate supplies of *Lachnosterna* grubs. An appeal to planters to find them and send them in had not produced sufficient response, and those sent in were usually confined in too small a receptacle, and were too bitten and damaged to be of any use for laboratory breeding work. At that period of the year, it was, moreover, not very easy to locate *Lachnosterna* areas; it was too early for fields of growing cane to show damage from *Lachnosterna* attack, particularly as rainfall had been sufficiently abundant to keep the canes green and healthy, and, unless a definite attack is evident, there is no excuse for digging up apparently healthy fields of canes to see if *Lachnosterna* grubs are there. The only alternative was to find grubs in thrown out cane fields which had not yet been stumped or ploughed. As these could give no surface indication of the presence of *Lachnosterna* grubs, considerable search was necessary before a worth while supply of brown hardback grubs was obtained: heavy rains also made the digging for grubs difficult.

Gradually, however, supplies were built up and stored in large tins of moist soil.

The breeding procedure followed was that used by Mr. Jepson and the writer in Puerto Rico based on Jarvis' method (5). Moistened sterilised soil was placed in wide-mouthed, cylindrical tins (Quaker Oats tins were used in Puerto Rico) to a depth of about one inch, and then two depressions were made in the soil on opposite sides of each tin and a grub placed in each, with a roof of slightly compressed soil over each grub. The tins were then filled to within

an inch of the top with soil lightly shaken or pressed down, and a fertile wasp placed on top of the soil with a leaf on which water and honey solution was put, and a well-ventilated lid placed on each tin.

Twenty-four hours later, each tin was opened up, and the wasp caught in a tube. The remaining soil was then removed, and the grubs recovered. If they had eggs deposited on them, they were placed each in a small separate tin of soil (ointment tin or 'Oxo' tin), dated, and allowed to develop. It was found in the preliminary work on the first consignment that mortality was very high when numbers of parasitised grubs were placed for development in a large shallow box with an adjustable glass lid. Each breeding tin was then replenished with grubs, refilled, and the wasp returned to it; every four or five days the wasps were placed in cages with food and males, and given twenty-four hours' rest.

By keeping each parasitised grub in a separate tin, the spread of mould or bacterial rot was prevented and individual records were easier to follow.

As a result of this breeding work, it was speedily apparent that *E. ephippium* was the only Scoliid from which any laboratory results could be expected.

C. tricineta, F. (*pyrura* Rohwer) either killed the *Lachnosterna* larvae by its sting, or, if eggs were laid, larvae did not develop; or, if larvae developed, they either wandered off their host or died. No cocoons at all were obtained.

C. trifasciata gave slightly better results. Oviposition was sparse and irregular, but the larvae hatched out and developed on their new host, and six ultimately pupated. From these, two males were obtained, which were put back to the few surviving females. No further development occurred, and this species died out.

E. haemorrhoidalis gave faint expectations of success in that up to September 26th, 9 cocoons had been obtained which yielded one female and two males. There was a tendency, however, for the females to kill the *Lachnosterna* larvae by their sting, and for the eggs, when laid, to become watery and fail to develop. This species also soon died out in the laboratory. The life cycle recorded for the few specimens obtained was 8 days from egg to pupa, and 40 days from pupa to adult, making a total life cycle of 48 days.

E. ephippium (*xanthonotus*), as has been stated, gave quite a promising result, and was reared through five generations in the laboratory, ending on August 23rd, 1934, with the last of a series of males hatching. The following Table I will show the numbers obtained in each generation and the proportion of males to females which gradually led to the cessation of laboratory breeding.

At one time in the second generation, it looked as if *ephippium* was adapting itself successfully to the new host and condition, and that sufficient numbers would eventually be available not only for liberation in Barbados, but for transmission to Mauritius for Mr. Jepson, it being part of the scheme of co-operation that, if any of the introduced Scoliids proved successful in Barbados in the host *L. smithi*, which is common to Barbados and Mauritius, supplies of the laboratory reared Scoliids would be sent to Mr. Jepson in Mauritius. Pre-

liminary arrangements were in fact made by letter to Capetown, South Africa, to ensure trans-shipment and care of the *Seciids*, should a shipment prove possible. A small consignment of fifteen first generation cocoons were also sent to Mr. Jepson in Puerto Rico as a nucleus of material bred on the new host *L. smithi*.

As shown by Table I, however, all hopes of increased laboratory supplies soon disappeared.

TABLE I.
LABORATORY RECORDS OF *E. EPHIPIUM* (*XANTHONOTUS*).

Generation	Dates of hatchings.	No. of pupae.	No. of males.	No. of females.	Total.	Percentage of females
First ..	13/10/33 to 4/12/33	85	53	12	65	18.4
Second ..	16/11/33 to 5/ 4/34	83	47	17	64	26.5
Third ..	2/ 2/34 to 28/ 7/34	111	58	11	69	15.9
Fourth ..	14/ 4/34 to 21/ 8/34	51	30	5	35	14.2
Fifth ..	4/ 7/34 to 23/ 8/34	23	18	1	19	5.2
Total ..		353	206	46	252	18.2

The periods of development were irregular, but in the first generation the average from egg to pupa worked out at 7 days, and from pupa to adult 40.7 days, giving a total cycle of 47.7 days. In the fifth generation, egg to pupa took 7.7 days and pupa to adult 41.1 days as an average, making a total cycle of 48.8 days.

It will also be seen from the above Table that the percentage of emergences from cocoons was around 76 per cent. in the first two generations, and then dropped to 62 per cent. in the third and most promising generation. This generation pupated and developed during the hot, dry season in Barbados, and, despite keeping the cocoons in plugged glass tubes on pads of cotton wool above an inch layer of moist sand, emergences of adults became so irregular that eventually cocoons had to be opened and the wasps assisted out when their normal period of development had elapsed but for this procedure, the number of wasps hatching in the third generation would have been still less. As it is, the third and fourth generations declined very rapidly in numbers and vitality, and the proportion of females became so low that the species became extinct in the laboratory.

At the time that these Scoliid were introduced, the favourite food plant of *ephippium* and *haemorrhoidalis*, namely *Mitracarpus portoricensis*, was introduced into Barbados, and, from plants grown at Codrington Experimental Station, sufficient seed was obtained to furnish a supply of young plants which it was hoped to plant on waysides and cart tracks when the rainy season began, which would, it was hoped, coincide with liberations of laboratory-reared *ephippium*. The rainy season, however, did not materialise in 1934, and, as shown, the artificial rearing of *ephippium* dwindled to vanishing point.

The success of the Scoliid introduction, therefore, depends upon the liberations made direct from material obtained from Puerto Rico, namely 174 males and females of all species combined, of which the majority were *E. ephippium* (*xanthonotus*) females.

Only *E. ephippium* proved adaptable to its new host *Lachnosterna smithi*, and, judging by laboratory records, there is not a strong possibility at all that even this species will have established itself in the field.

Whilst in Puerto Rico, Mr. Jepson also studied the Tachinid fly *Cryptomeigenia aurifacies*, Walton, which is parasitic on *Melolonthid* beetles in the northern and more humid areas of Puerto Rico, but the extent of parasitism was found to be so small that no attempt was made by the writer to introduce it into Barbados.

The next introduction to be considered is that of *Pyrophorus luminosus*, the larvae of which are predatory on white grubs in Puerto Rico. Dr. G. Wolcott of the Experiment Station, Rio Peidras, Puerto Rico, had been approached in 1932 concerning the status of *P. luminosus* (known in Puerto Rico as Cucubano) and the possibility of obtaining material for shipment to Barbados. Dr. Wolcott mentioned this to Mr. W. F. Jepson on his arrival in Puerto Rico, and as a result, Mr. Jepson very kindly sent about 1,200 adult *P. luminosus* beetles, collected at Cidra towards the end of May in the hillier region towards the centre of Puerto Rico. Unfortunately, the percentage of females to males collected in the field is very low, and Mr. Jepson had found by dissection that, out of forty beetles so captured, there were three females only; it is probable, therefore, that a similarly low percentage of females was present in the above sending.

These beetles came by air mail to St. Lucia, whence they were delivered in Barbados.

The consignment arrived in good condition, with very few beetles dead, and half were liberated at Blackmans Plantation, St. Joseph, and half at Cane Garden, St. Thomas, where it was considered that the beetles would have the maximum chance of suitable climatic conditions and abundance of hosts.

No searches have been made to discover whether the beetle has established itself, because it is considered that if it does survive and flourish, the appearance of 'fireflies' will speedily be reported.

The writer wishes to take this opportunity of expressing his thanks to Mr. Jepson for the sendings of parasites which have been recorded in the foregoing pages, and for his willing and effective co-operation.

Mr. Jepson's preliminary surveys, and establishment of working headquarters in the field, enabled considerable ground to be covered during the writer's stay of 30 days in Puerto Rico.

The project of introducing Cucubano to Barbados did not cease with the above consignment. Searches were made for the larvæ in the field in Puerto Rico during the writer's visit in June to July, 1933, but, owing presumably to the main emergence and mating flight of beetles being in May, it is usually not until ploughing season in September onwards that Cucubano larvæ can be found in sufficient numbers. In September, 1933, Mr. Jepson found Cucubano larvæ in considerable numbers at Cidra.

Negotiations were therefore started with Dr. Wolcott for a similar supply for Barbados to be sent in 1934. A trial sending of a Cucubano larva, apparently quite one third grown was made by ordinary post by Dr. Wolcott at the end of December, 1933. This larva, which was sent in an ointment tin, plus a little coir, arrived in sound condition and has flourished in a jar of soil for 24 months in Barbados; it consumed at least one *L. smithi* larva per day.

Dr. Wolcott's attempts to find a suitable supply of Cucubano larvæ in the autumn of 1934 were not successful, and at the beginning of December, 1934, only one consignment of 12 larvæ had been received in Barbados, of which one was dead and two others subsequently died. The remaining 9 were placed out in a field at Ayshford, St. Thomas, which was heavily infested with *L. smithi* larvæ.

Early in 1935, however, large consignments of Cucubano larvae were received from Dr. Wolcott, and during January, February and March of 1935, a total of 3,670 were placed out in infested fields at Ayshford, Hopewell and Walkes Spring, in the Parish of St. Thomas.

On the writer's return to Barbados from England in November, 1935, enquiries elicited the information that considerable numbers of 'fire-fly' beetles had been seen in May at Hopewell near the field in which the larvæ had been placed, and that some had been seen at Ayshford. It was also reported that Cucubano larvæ had been dug up subsequent to May in the previously infested fields, in which no *Lachnosterna* larvæ could be found.

The emergence of beetles in May corresponds with the emergence time in Puerto Rico, and such specimens undoubtedly came from the large, nearly mature larvae placed in the ground. The Cucubano larvae found in the soil would be those which were immature when placed in infested fields in 1935, and would not be due to emerge until May of 1936. This is also borne out by the fact that two immature Cucubano larvae kept for observation are quite half grown, and still alive in their jars, despite the fact that they have had no food whatsoever for nearly nine months. The original Cucubano larva, sent by post in December 1933, is also still alive in its jar in the laboratory, and appears healthy, though it has also been without food for nearly nine months.

It appears, therefore, that the larvae are hardy, and will live and feed either in jars in the laboratory, or in the field, and that numbers of adult Cucubano have emerged in Barbados in May, 1935, and that others should emerge in May, 1936. As to whether these adults have bred successfully and will maintain a population

of Cucubano larvae in the soil of the hillier, wetter areas, can only be shown by time. The introduction, however, gives promise of some measure of success if each larva lives for one or two years in the soil, and eats or destroys one *Lachnosterna* larva per day.

Before leaving the subject of biological control of the brown hardback in Barbados, some reference must be made to the toad *Bufo marinus*. The giant toad exists in Barbados, and is known to be useful on account of its insectivorous diet. Its value as a control over *L. smithi*, or other beetles of this nature, has recently become very apparent in Puerto Rico, into which country it has been introduced in recent years from Barbados and from Jamaica.

In Puerto Rico, the giant toad has found very favourable conditions for breeding and can now be found in large numbers everywhere, particularly in the irrigated Southern cane growing areas, where May beetles (brown hardbacks) and root borer beetles (*Diaprepes abbreviatus*) were at one time serious pests.

The writer found on his visit to Puerto Rico in 1933, that both the above pests were no longer considered to be serious on the sugar lands which served the larger Southern Centrales, and the reduction in these pests, particularly the brown hardbacks, or Caculos, as they are called there, was ascribed entirely to the large numbers of *Bufo marinus* toads, and to their voracious feeding on the adult and probably newly emerged beetles. This view is emphatically upheld by Dr. G. N. Wolcott who was satisfied himself that the giant toad is an effective controlling agent over the pests in question.

Unfortunately, in Barbados conditions are not so favourable for the rapid increase of this toad, for there is very little surface water or permanent natural shelter in existence at all and any permanent water is invariably oiled against mosquitoes.

According to Schomburgk, *Bufo marinus* was introduced into Barbados, presumably from British Guiana, about 1830, and was, in 1848, present all over Barbados in large numbers. The fact that it is not present in large numbers now can be ascribed mainly to the disappearance of surface water and natural shelter. Thus, H. A. Ballou, (1), states "According to this observer (Dr. C. J. Manning, in Education Gazette, August, 1914) the planters of Barbados have completely changed the physical aspect and conditions of the Island during the past 50 years. Formerly, each of the 400 and more estates in the Island had at least one pond, and most of them had more, and it is estimated that there were over 1,000 ponds, all brimfull of water during the wet season. Now the ponds have all been drained, many pastures have been planted, ravines and gullies have been very much cleaned, so that they provide for a quick run off of the water. The trees have been cut down to a very large extent."

This lack of water for breeding, and diminution of natural shelter, has caused the toads to diminish in numbers to the point of ineffectiveness. It is considered by Ballou that the mongoose is not a serious contributory cause to the diminution because the poisonous secretion of the toad would be an effective protection.

Many planters in Barbados ascribe the increase in brown hardback and root borers to the diminution in the numbers of giant toads, and to the disappearance of frogs and lizards in general.

There appears to be some support for this idea in that, as stated above, the recent introduction and rapid increase of *B. marinus* into Puerto Rico is reported to be responsible for the decrease in damage of the cane root pests which appear to have increased in Barbados with the decrease in numbers of the toad.

Biological control need not be restricted to insects versus insects, and it appears to the writer that there is a strong case for the artificial propagation and protection of *B. marinus* in Barbados, in order to restore it to its one time status.

If it is found possible and practicable to increase and maintain its numbers artificially, there is no doubt that this toad is capable of dealing very effectively with *Lachnosterna* and *Diaprepes* when they emerge fresh from the soil, and particularly with *Lachnosterna* which returns again to the soil for shelter in daytime and for egg-laying purposes. The toad also has its uses against cotton and sweet potato defoliators.

The artificial breeding and distribution of *B. marinus* in Barbados needs considerable fundamental investigation, but, in the light of recent knowledge, it appears both practicable and essential as a control measure against brown hard-backs and root borers.

IV. Control of *Pectinophora gossypiella*, Saund.

The introduction of *Microbracon kirkpatricki* from Uganda, to control the pink bollworm in cotton in Barbados, was undertaken in May, 1929, through the kind assistance of Sir Guy Marshall of the Imperial Institute of Entomology and Dr. W. R. Thompson of the Farnham Royal Laboratory.

A consignment of these parasites was received from Dr. Thompson on May 15th, 1929, in glass tubes, the corks of which had split raisins fixed inside the tube as food for the parasites.

On being opened up, 62 were dead and 31 alive; and, of the latter, 16 were females and 15 were males.

As May is the close season for cotton in Barbados, there was no field infestation on which to draw for supplies, but green cotton bolls were obtained from a few plants grown in cages at the Experimental Station for breeding work, and a small supply of pink bollworm was obtained from vessels which brought cotton seed to Barbados. This seed is brought for oil extraction, and is sterilised in a Simon's Heater before being brought ashore, and the vessels cleaned up and the holds heavily fumigated with Hydrocyanic acid gas. Prior to these operations, the larvae required for experimental work were taken out.

These larvae, as obtained, were placed in the green bolls as follows: the tops of the bolls were cut off and some seeds removed from one loculus: a pink bollworm was then inserted, next to the skin of the boll, and the seeds, slightly crushed, replaced so as to hold the larva in position.

These prepared bolls were then exposed to the parasites: in some cases the boll was pierced at the spot where the larva was to be inserted, to facilitate oviposition on the part of the parasites.

The parasites were kept supplied daily with food and with freshly prepared bolls, and survived for twelve days in the laboratory.

From the majority of bolls so exposed, pink bollworm moths only were recovered, and no parasitism by *Microbracon kirkpatricki* had occurred, though dissection of the bolls showed that the pink bollworm had, in most cases, pupated next to the skin of the boll, in an ideal position for parasitism through the boll wall.

A second introduction of this parasite in 1930 was diverted to Trinidad, on account of the writer's absence in America at that period. Dr. J. G. Myers handled the consignment, and obtained, in the first generation males only, so that this introduction failed. A report, received in 1930 from El Paso, Texas (F. A. Fenton and L. W. Noble), also showed that the breeding of this introduced parasite was unsuccessful there.

This biological project was therefore abandoned.

V. Control of Lepidopterous Defoliators.

The introduction into Barbados of the Tachinid *Compsilura concinnata*, Meigen, from the Gipsy Moth Laboratory, U.S.A., was suggested to the writer by Dr. W. R. Thompson at Farnham Royal Laboratory in 1930.

This Tachinid, a native of Europe introduced into the United States, has a wide range of hosts in New England, U.S.A., as indicated by Departmental Bulletin 1363 (8) and has been used with considerable success in the control of the Gipsy Moth. There was a reasonable possibility, therefore, of its finding a suitable host or hosts in the various lepidopterous larvae such as species of *Cirphis*, *Euthiasonata*, *Papilio*, *Anosia*, *Vanessa* and *Sphingid* larvae which occur in Barbades, and which are known to be parasitised in other countries by *C. concinnata*.

It was hoped that at the same time it might attack the larvae of species of *Feltia*, *Heliothis*, *Laphygma*, *Mocis*, *Prodenia*, *Xyloxyges*, *Phytometra*, *Thermesia* and other lepidopterous pests which attack economic crops, particularly sweet potato, corn, cotton, and cover crops.

In September, 1930, therefore, the U.S. Bureau of Entomology was approached with a view to a shipment of *C. concinnata* from the Melrose Highlands Laboratory, Massachusetts, with the result that in July, 1931, a consignment of 1,000 puparia was received from Dr. C. W. Collins, from the Gipsy Moth Research Laboratory.

Unfortunately, the cold storage conditions on board ship during transit could not have been too good, for the contents of the package containing the *Compsilura* puparia had obviously heated up *en route*. When the container packed with damp sphagnum and puparia was opened up, a large number of flies had already hatched out and were dead, whilst a few were alive and others were hatching. Altogether 27 healthy flies were obtained twenty-four hours after receipt of the consignment.

All remaining sound puparia were sorted out, mounted on cards, and placed in gauze capped glass tubes for hatching and observation for hyperparasites: the remaining material was destroyed.

No more flies were obtained after the first twenty-four hours, but a few hyperparasites subsequently hatched out.

The adult flies were placed in a large 'Vita-glass' cage with a supply of various species of lepidopterous larvae, and one end of the cage was exposed to diffused sunlight. Observation did not show any attempts at mating or of parasitism of the larvae, and, as 8 flies soon died, the remaining 19 were placed out in a large field infested with larvae, mainly *Thermesia* (*Anticarsia*) *gemmatilis*: other lepidopterous larvae were also present to a less extent in the same and neighbouring fields.

In June, 1932, Dr. C. W. Collins made a further shipment of over 1,400 puparia of *C. concinnata* which was received in Barbados in the following month.

The writer was then in England on leave. However, this time the cold storage arrangements in transit had been satisfactory, and a total of 1,205 flies hatched after arrival in Barbados, from a total of 1,432 puparia received. The majority of flies hatched on the 5th, 6th and 7th days after arrival, and hatching ceased on the 11th day.

No hyperparasites emerged, and a fortnight after the last emergences, the material was destroyed by burning.

Liberations were made as follows: 82 on July 16th and 306 on July 17th at Codrington Experimental Station: 437 on July 18th at Halton, St. Philip, and 362 on July 19th at Codrington Experimental Station. Several pairs were seen mating.

No field recoveries have yet been made, but it is hoped that if the parasite has established itself, it will gradually appear in sufficient numbers to render its recovery inevitable.

SUMMARY.

An account is given of fourteen insect parasites introduced into Barbados since 1927 in an endeavour to secure biological control over insect pests of various categories.

No positive results have been obtained from the majority of these introductions. Some projects, such as the establishment of *Lixophaga diatraca* and *Pyrophorus luminosus* are still in progress, and one parasite, namely, *Paratheresia claripalpis* is to be re-introduced.

The status of *Bufo marinus*, the giant toad introduced over 100 years ago into Barbados, is discussed: the reasons for its diminution to the point of ineffectiveness noted and suggestions made for its artificial propagation as a control over the pests *Lachnosterna smithi* and *Diaprepes abbreviatus*.

REFERENCES.

- (1) Ballou, H. A. .. The Toad in the West Indies. The Agricultural News. Vol. XVIII, No. 459, Nov., 1919, p. 378.
- (2) Box, H. E. .. The Introduction of Braconid Parasites of *Diatraea saccharalis*, Fabr., into Certain of the West Indian Islands. Bull. Ent. Res. XVIII, Pt. 4, May, 1928.
- (3) Flanders, S. E. .. The Temperature Relationships of *Trichogramma minutum* as a basis for Racial Segregation. Hilgardia Vol. 5, No. 12. March, 1931, Berkeley, Calif.
- (4) Harland, S. C., & Atteck, O. M. .. Breeding Experiments with Biological Races of *Trichogramma minutum* in the West Indies. Zeitschrift fur inductive Abstammings und Vererbungslehre 1933. Bl. LXIV, Heft 1/2. Berlin.
- (5) Jarvis, E. .. The Biological Control of Cane Grubs. Tropical Agriculture, Vol. IX, No. 11, p. 331, 8 figs., Trinidad, 1932.
- (6) Salt, G. .. Experimental Studies in Insect Parasitism. II. Superparasitism. Proc. Royal Society B. Vol. 114, 1934.
- (7) Thorpe, W. H. .. Biological Races in Insects and Their Significance in Evolution. Annals of Applied Biology. Vol. XXIII, No. 3, Aug., 1931.
- (8) Webber, R. T., & Schaffner, J. V. .. Host Relations of *Compsilura concinnata*, Meigen, an Important Tachinid Parasite of the Gypsy Moth and Brown Tail Moth. U.S. Dept. of Agriculture, Bulletin No. 1363, Washington, 1926.

**INVESTIGATIONS INTO THE USE OF THE ZEISS
HAND REFRACTOMETER.**

**(1) The Use of the Zeiss Hand Refractometer In the Early Stages of
Sugar Cane Seedling Selection.**

BY

A. E. S. McINTOSH, Geneticist,

AND

**G. C. STEVENSON, Assistant Botanist,
British West Indies Central Sugar Cane Breeding Station.**

INVESTIGATIONS INTO THE USE OF THE ZEISS HAND REFRACTOMETER.

(1) The Use of the Zeiss Hand Refractometer In The Early Stages of
Sugar Cane Seedling Selection.

TABLE OF CONTENTS.

	PAGE.
I. INTRODUCTION	25
II. MATERIAL AND METHODS	25
III. PRESENTATION OF RESULTS	28
IV. DISCUSSION	33
V. CONCLUSIONS	36
VI. FUTURE LINES OF WORK	37
VII. SUMMARY	37
VIII. REFERENCES TO LITERATURE	39

I. Introduction.

Experiments on the use of the Zeiss Hand Refractometer in seedling selection work have been carried out by Verret and Mangelsdorf in Hawaii (7), and Craig in Mauritius (1, 2). These investigators have shown that this instrument provides an easy and reliable means of determining the comparative juice qualities of seedlings. The Zeiss Hand Refractometer determines the refractometric brix of seedlings, and this has been shown to bear a high positive correlation to the sucrose in juice as indicated by the polariscope.

The advantage of the Zeiss Hand Refractometer in this work lies in the ease and rapidity with which it can be used in the field, thus avoiding the necessity and expense of transporting samples of cane to the laboratory for routine milling and sucrose determinations.

In the work of seedling raising and selecting in Barbados, the first two seasons of test (i.e. the first and second year seedling trials) are chiefly devoted to a selection of seedlings of approved general standard. It has been shown elsewhere that considerable importance in selection work is attached to the sucrose per cent.-in-juice figure (4). Moreover, in the first year seedling trial, populations of crosses are compared, among other features, on the extent of sucrose per cent. in juice of their seedlings. The object of this is to determine the value of individual parents and crosses in endowing juice quality to their progeny.

Up to the present, the estimation of the sucrose per cent. in juice of individual seedlings and populations of seedlings has been made from the juice derived from milled cane samples. This involves considerable labour and expense and also limits the number of samples, and so seedlings, whose sucrose in juice can be determined.

During the crop of 1935, experiments were carried out to find out (1) the most reliable field sampling method and (2) the degree of usefulness of the Zeiss Hand Refractometer in indicating the sucrose in juice of seedlings. The latter was determined by correlating Refractometer readings with sucrose in juice percentages established by laboratory methods, the former by varying the sampling technique and correlating the results of each sampling method with the laboratory results.

II. Material and Methods.

(a) *Material.*

The seedlings employed in the investigation were those selected in the field during the reaping of the first year seedlings, and those reaped at the second year seedling trials, of the 1935 crop.

The methods of conducting first year and second year trials in Barbados have been reported in detail elsewhere (5).

In the *first year seedlings*, there were approximately 6,500 in the Proven Cross Group* which were reaped at two times—early (February) and late (April).

*This group provides selections of potential commercial value for *thick-cane* areas in the British West Indies. In 1935, it contained seedlings derived from nine noble cane crosses.

Refractometer brix readings were made for 234 seedlings at the early reaping and 204 seedlings at the late reaping.

In addition to the above, there were approximately 3,500 seedlings in the Experimental Cross Group** which were reaped in March. These seedlings were the progeny of approximately thirty crosses, whose parent material represented a wide range in origin. Refractometer readings were taken for 172 seedlings in this group.

The *second year seedling trials**** were reaped at two stations—low rainfall and high rainfall—in March and May respectively. They included thirteen seedlings and two standard varieties, planted in four randomised blocks. At Dodds (low rainfall station), the fifteen varieties were sampled in two blocks. At Todds (high rainfall station), the thirteen seedlings only were sampled, in each of the four blocks.

(b) *Methods.*

Refractometer brix determinations were made in the field in the following manner. A few drops of juice were obtained by thrusting into the cane a metal gouge similar to that described and illustrated by Levert, van der Wonde and van Dillewijn (3). This was done by a labourer, working under the supervision of an officer. The latter read the brix from the scale of the instrument, afterwards removing the juice with a damp cloth and drying the refractometer. A second officer recorded the determinations. Zero readings were taken to check the instrument in the morning, at midday, and at the conclusion of the day's work. It is of interest to note that no alteration of the zero adjustment was necessary during the whole of the season's work.

It was possible, using the personnel indicated above, to make approximately a thousand brix determinations per day, reading the scale to 0.1 per cent. brix with little difficulty.

Preliminary experiments, designed to test the working and accuracy of the refractometer, and to indicate possible effective sampling methods, showed that (1), duplicate readings from the same internode of a cane were quite unnecessary in sampling a seedling. It was clear from the close approximation of such duplicate readings that the sampling error of the internode was very low; (2) in the stool of any one variety canes of any one age class—three classes being made and arbitrarily designated 'a', 'b' and 'c' canes—gave approximately similar readings when sampled internode by internode; (3), readings from corresponding positions in canes of different age classes differed appreciably in immature stools, although this difference was much less marked in mature stools; (4) samples taken along the cane showed appreciable differences, although here again such differences were less marked in mature canes.

Following these preliminary experiments, the sampling schemes for the first year and second year seedlings were worked out. These are described below.

**This group serves to test the value of parent varieties as seedling producers, and to provide varieties suitable for growing commercially under *special* conditions in the British West Indies.

***These trials serve solely to test seedlings of potential commercial value for *thick-cane* areas in the British West Indies.

First Year Seedlings. The cut canes of each selected seedling were examined, and four canes chosen; age types being represented approximately in the ratio in which they occurred in the stool. From each of the four canes, a brix sample was taken from each of the middle of three internodes. The internodes were chosen at about one foot from the base, at the middle, and at about one foot from the cut top. These are called for convenience, bottom, middle and top respectively. In taking the samples with the gouge, only healthy internodes, free from moth borer or other damage, were chosen. A duplicate sample, from the same or from the adjoining internode, was sometimes taken, if the reading appeared to be abnormal. In such cases, moth borer damage or rotting was generally found to be the cause of the abnormality. In order to determine the simplest and most effective sampling method,—that is, the method which gives the closest indication of the sucrose in juice of the seedlings—the readings were combined in various ways. The mean brixes of each and every combination were correlated with the appropriate sucrose percentages. The combinations tested are noted below in Table I.

TABLE I.
FIRST YEAR SEEDLINGS.

SCHEME OF COMBINING SAMPLE REFRACTOMETER READINGS OF INDIVIDUAL SEEDLINGS.

Sampling Combina- tion.	Description.	No. of Sam- ples to give mean.
1	Middle positions from two canes ..	2
2	Bottom " " " " ..	2
3	Middle " " three " ..	3
4	Bottom " " " " ..	3
5	Bottom & Middle " " two " ..	4
6	Middle " " four " ..	4
7	Bottom " " " " ..	4
8	Top, Middle & Bottom " " two " ..	6
9	Middle & Bottom " " three " ..	6
10	" " " " " four " ..	8
11	Top, Middle & Bottom " " three " ..	9
12	" " " " " four " ..	12

In cases of establishing mean brixes with readings from the records of two or three of the four canes only, the readings from the majority cane age-class were employed.

Second Year Seedlings. The second year seedlings were randomised in four blocks, the individual plot containing twenty-four stools. At reaping, the usual sample bundles of 80 lb. from each plot were set aside for laboratory analysis.

In making refractometer brix determinations, seven canes were selected at random from the laboratory bundles, and each cane was sampled at three positions, as for the first year seedlings described above. Sucrose in juice figures were obtained for each bundle.

III. Presentation of Results.

First Year Seedlings. The mean refractometer brix for each seedling for each of the twelve combinations of readings noted in Table I was ascertained.

Target diagrams were prepared, showing the relationships between the brix and sucrose percentage means of the individual seedlings. This gave twelve target diagrams for each group. Correlation coefficients were determined from each of the twelve diagrams in the early and late proven cross groups, and the experimental cross group. These coefficients are noted in Table II below.

TABLE II.

First Year Seedlings—1935.

SHOWING CORRELATION COEFFICIENTS BETWEEN MEAN REFRACTOMETER BRIX OF
VARIOUS SAMPLING COMBINATIONS AND SUCROSE PER CENT. IN JUICE.

A. *Proven Crosses — Early Group.* 234 Seedlings.

Sampling * Combination.	Average of Means of Refractometer Brix Readings.	Mean Sucrose Per Cent.	Correlation Coefficient.	Significance. (Probability —P.)
1	19.28	14.62	+ 0.7795	< .01
2	19.60	14.62	+ 0.7775	< .01
3	19.28	14.62	+ 0.8140	< .01
4	19.52	14.62	+ 0.8184	< .01
5	19.43	14.62	+ 0.8180	< .01
6	19.23	14.62	+ 0.8127	< .01
7	19.44	14.62	+ 0.8261	< .01
8	19.07	14.62	+ 0.8719	< .01
9	19.44	14.62	+ 0.8436	< .01
10	19.30	14.62	+ 0.8392	< .01
11	18.93	14.62	+ 0.8971	< .01
12	18.88	14.62	+ 0.8763	< .01

* C.f. Table I.

TABLE II.—*Continued.***First Year Seedlings—1935.**

SHOWING CORRELATION COEFFICIENTS BETWEEN MEAN REFRACTOMETER BRIX OF
VARIOUS SAMPLING COMBINATIONS AND SUCROSE PER CENT. IN JUICE.

B. Proven Crosses — Late Group — 204 Seedlings.

Sampling Combination.	Average of Means of Refractometer Brix Readings.	Mean Sucrose Per Cent.	Correlation Coefficient.	Significance. (Probability —P.)
1	21.47	17.12	+ 0.7735	< .01
2	21.10	17.12	+ 0.7631	< .01
3	21.06	17.12	+ 0.8058	< .01
4	21.17	17.12	+ 0.7128	< .01
5	21.24	17.12	+ 0.8045	< .01
6	21.43	17.12	+ 0.8376	< .01
7	21.28	17.12	+ 0.7750	< .01
8	21.35	17.12	+ 0.8081	< .01
9	21.22	17.12	+ 0.8248	< .01
10	21.36	17.12	+ 0.8081	< .01
11	21.36	17.12	+ 0.8349	< .01
12	21.49	17.12	+ 0.8566	< .01

TABLE II.—*Concluded.***First Year Seedlings—1935.**

SHOWING CORRELATION COEFFICIENTS BETWEEN MEAN REFRACTOMETER BRIX OF
VARIOUS SAMPLING COMBINATIONS AND SUCROSE PER CENT. IN JUICE.

C. Experimental Crosses — 172 Seedlings.

Sampling Combination.	Average of Means of Refractometer Brix Readings.	Mean Sucrose Per Cent.	Correlation Coefficient.	Significance. (Probability —P.)
1	19.80	15.12	+ 0.7181	< .01
2	20.46	15.12	+ 0.6680	< .01
3	19.72	15.12	+ 0.7183	< .01
4	20.45	15.12	+ 0.6837	< .01
5	20.19	15.12	+ 0.7353	< .01
6	19.70	15.12	+ 0.8170	< .01
7	20.41	15.12	+ 0.7818	< .01
8	19.96	15.12	+ 0.7592	< .01
9	20.11	15.12	+ 0.7579	< .01
10	20.10	15.12	+ 0.7852	< .01
11	19.76	15.12	+ 0.7609	< .01
12	19.76	15.12	+ 0.8578	< .01

These various correlations are illustrated diagrammatically in Figure I.

A specimen target diagram for one sampling combination in the early proven cross group is illustrated in Figure II.

Second Year Seedlings. The refractometer brix of each sampled cane was ascertained by taking the mean of its three samples. The refractometer brix of the laboratory field sample bundle was determined by taking the average of the means of (1), three canes and (2), seven canes; this in order to find out if it was necessary, for appreciably better correlations, to sample more than three canes. Correlations were determined between the brix averages of the means of three and of seven canes and appropriate sucrose per cent. in juice figures. These correlations regarded the individual plot sample as the unit.

In addition, the seedling was regarded as unit and correlations determined between seedling brix means—for each seedling, the mean of two plots at Dodds and four plots at Todds—and seedling sucrose percentages in juice.

Target diagrams were prepared for each of these correlations.

The coefficients of correlation are noted in Table III.

TABLE III.

Second Year Seedlings.

COEFFICIENTS OF CORRELATION BETWEEN VARIOUS MEAN BRIX DETERMINATIONS
AND SUCROSE PERCENTAGES.

Place	Correlation	Numbers Correlated	Correlation Coefficient.	Significance (Probabil- ity — P)
Dodds ..	} Plot Refractometer Brix derived from	30	+ 0.8005	< .01
Todds ..				
	} three canes and Sucrose Per Cent.	52	+ 0.7364	< .01
Dodds ..	} Plot Refractometer Brix derived from	30	+ 0.8443	< .01
Todds ..				
	} seven canes and Sucrose Per Cent.	52	+ 0.8438	< .01
Dodds ..	} Seedling Refractometer Brix and	15	+ 0.8024	< .01
Todds ..				
	} Sucrose Per Cent.	13	+ 0.8526	< .01

FIG I.

Correlation between Hand Refractometer
Brix and Sucrose per cent in Juice.

Line Graphs showing comparative
Correlation Coefficient values
for various Field Sampling Techniques.

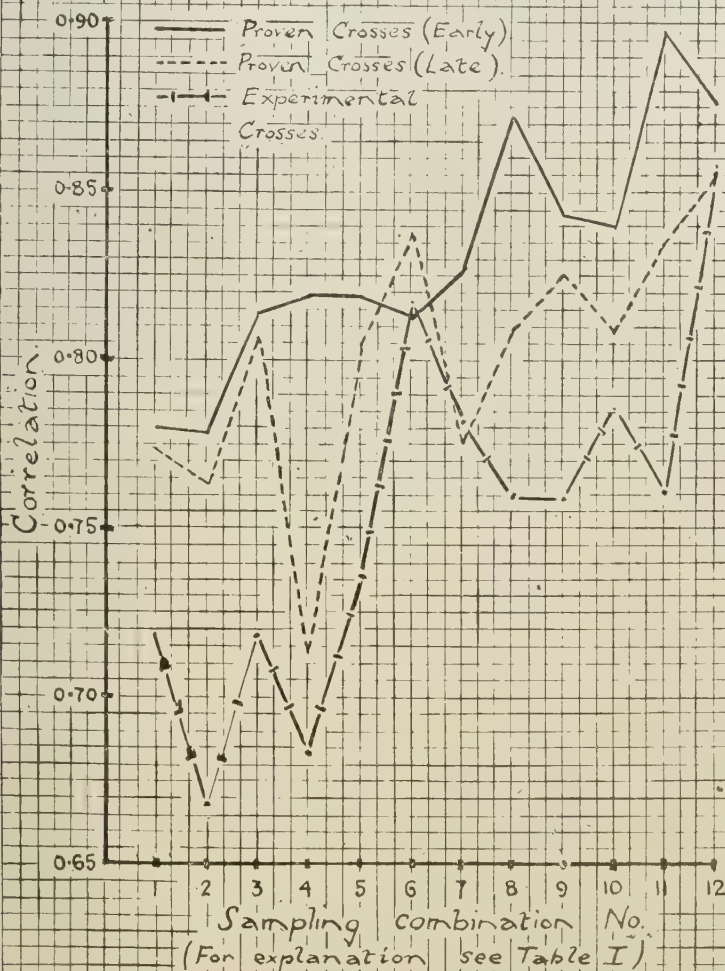
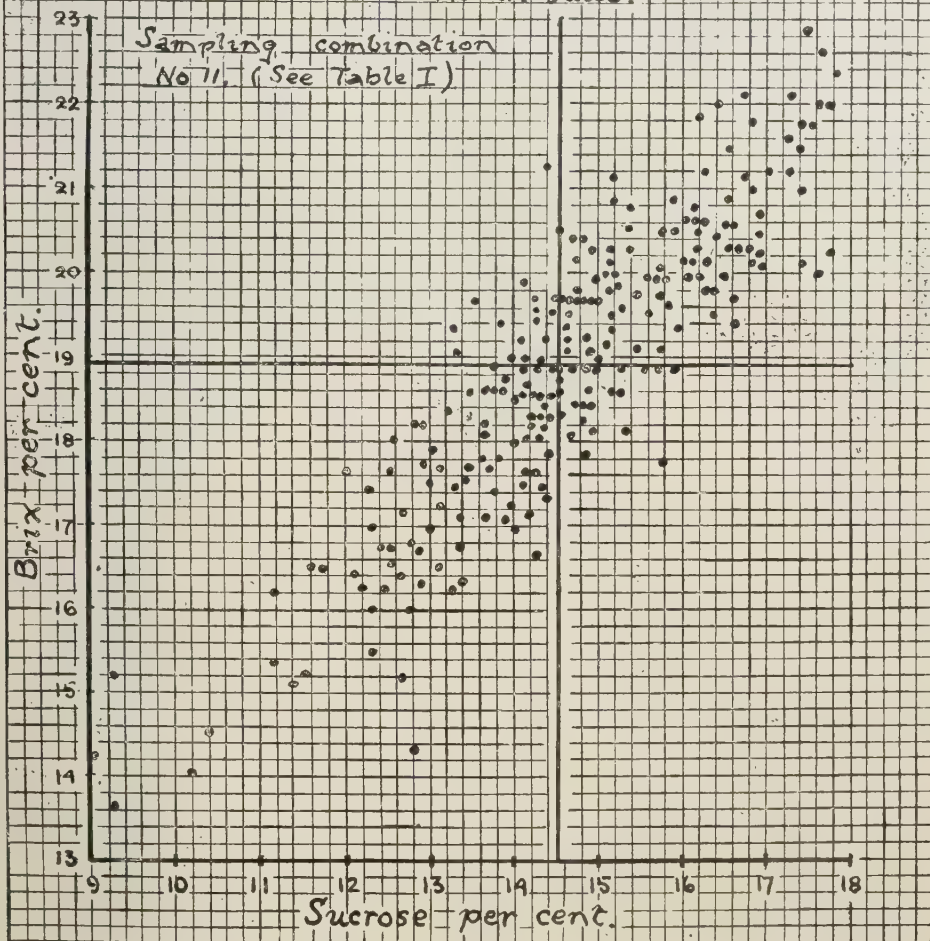


FIG II.

FIRST YEAR SEEDLINGS ~ EARLY.
PROVEN CROSSES.

Target Diagram showing the relationship between
Hand Refractometer Brix and Sucrose per
cent in Juice.



IV. Discussion.

The per cent. sucrose in juice is obviously a highly important commercial characteristic in sugar cane seedlings. It is therefore important that it be used in seedling selection work, if it is known to persist in extent throughout generations derived from vegetative propagation. It has been conclusively proved in Barbados (6) that batches of seedlings in successive generations, and under conditions rendering legitimate a comparison of sucrose per cent. in juice, develop this feature to a remarkably constant comparative degree.

In later stages of seedling selection work, it is in addition necessary to make complete chemical analyses of seedlings. Sucrose determinations alone, therefore, apply to seedlings in the early stages of selection. This, in Barbados, is confined to the first and second year seedling trials.

Sucrose determinations involve appreciable labour and expense, more particularly where large numbers of seedlings are being dealt with: e.g. in the first year seedling trial. Any reasonably accurate method which would lessen this work and expense is evidently justifiable.

The experiments noted in the two previous sections would appear to prove that, by certain methods, the Zeiss Hand Refractometer is a very reliable and simple instrument in indicating the sucrose per cent. in juice.

The correlations established deal essentially with seedling populations, the units employed being, in the case of first year seedlings, samples from individual stools, in second year seedlings, samples from single plots and seedling means derived from groups of plots.

In all cases the correlation coefficients were high and definitely significant.

Several features of interest are apparent in Table II with its various sections.

The following observations are made with regard to the sampling technique employed in first year seedlings. The two sampling variables were (1), number of canes sampled and (2), position of samples in individual canes. Each of these is seen to have a definite effect on the strength of the correlation. The time of reaping and the nature of the seedlings sampled also definitely affect the readings, and the correlations particularly, in so far as position of sampling in the cane is concerned. The effects of these factors are considered here.

The number of canes sampled. In the case of single samples from canes, it would appear that, in general, there is an improved correlation as the number of canes sampled is increased. This is particularly marked in late reaped seedlings and experimental cross seedlings, although not pronounced in early reaped seedlings. This may be explained by the fact that there exists a wider range of purities in a population derived from experimental crosses and late reaped proven cross seedlings. The former is doubtless due to a wider range in genotypes present in experimental cross seedlings, the latter to different rates of inversion in over ripe proven cross seedlings.

In the case of two samples from each cane, there is an improvement in the correlation derived from samples from three canes over two canes. There is practically no difference, however, between correlations derived from samples from three and four canes.

In the case of three samples from each cane, the use of three canes is slightly better than two, and four is only better than three in the case of experimental cross seedlings.

Position of Samples in the Cane. Four individual cane sampling methods were employed, i.e. (1), bottom alone, (2), middle alone, (3), middle plus bottom, (4) top, plus middle, plus bottom.

Where two canes were sampled, middle samples alone were slightly better than bottom samples in proven cross seedlings, although appreciably in experimental cross seedlings. The means from bottom plus middle samples were better than from middle and bottom samples alone. The introduction of the top sample effected an appreciable improvement in the correlation, particularly in the early group. This is possibly associated with inequality of ripening between seedlings in the early stages being most marked in the top (younger) internodes, and sampling these internodes in the early group gives a truer indication of the sucrose.

Where three canes were sampled, it is seen that bottom samples alone gave an appreciably poorer correlation than middle samples alone in the later reaped groups. This is probably associated with inversion being more marked in the lower (older) part of the cane and seedlings varying in their rates of inversion. The introduction of the middle and top samples tends to improve the correlation, more especially in the early group.

Where four canes were sampled, middles alone were appreciably better than bottoms in the experimental and late proven cross seedlings, although not so good in the early proven cross seedlings. The means of middle plus bottom do not give such a good correlation as the means of top, middle and bottom, although both are better in general than bottoms and middles alone.

The time of reaping. The early proven crosses, the experimental crosses, and the late proven crosses were reaped during early, mid—and late crop respectively. They may be said to have been unripe, ripe and, perhaps, overripe, respectively.

The effect of time of reaping on maturity would appear to have a marked result on the extent of the correlations, chiefly in so far as position of samples on cane was concerned. From the graphs in Fig I, it may be seen that the use of bottom samples was detrimental to the correlations in the experimental and late proven crosses. These are regarded as ripe and, possibly, in the case of many seedlings in them, over-ripe and inverting. The process of inversion need not alter the Zeiss Hand Refractometer Readings, although it has a decided effect on sucrose content.

The bottom or older portions of individual canes would naturally invert earlier, and it is reasonable to conclude that a differential rate of inversion in

seedling populations occurs and thus the inclusion of bottom samples, where inversion is most rapid, would tend to lessen the correlation coefficient.

The use of top samples improved the correlation in early reaped seedlings, i.e. seedlings not fully ripe. This is probably due to the use of an additional sample in each cane increasing the accuracy of the mean sampling figure of the cane.

The nature of the seedlings. Two categories of seedlings were tested in the first year seedlings, i.e. (1), proven crosses and (2), experimental crosses. It is certain that the range of genotypes is greater in the latter than in the former. It seems reasonable to conclude from this that a population of seedlings from the experimental crosses would provide a greater range in purities and in time of juice ripening and inversion. This in turn would lead to poorer correlations in a population of such seedlings. That this is so, is seen by a comparison of the correlation coefficients in Table II. (cf. Sections A and B with Section C).

It is of interest to note for the various groups the actual refractometer brixes and sucrose percentages, and the relationships between them. (See Table II Sections A, B and C.)

A comparison of the relationships between Zeiss Hand Refractometer brix and sucrose per cent. in juice between the proven crosses and the experimental crosses, would suggest lower purities in the latter.

In the second year seedlings, the sampling technique for individual canes consisted in taking juice samples from top, middle and bottom. A total of seven canes in the laboratory bundle was sampled. The correlations between means of three and seven sampled canes were established. The results showed that three sampled canes gave satisfactory correlations. The slightly better correlations obtained for seven sampled canes would not appear to justify the extra labour involved.

The actual selection practice, and in so far as sucrose in juice is concerned, is to take sucrose samples from each of the four plots and, for purposes of comparing seedlings, to compare the means of the four plots. (The evident use of four replicates is to attempt to overcome the effect of soil heterogeneity on juice quality).

The Zeiss Hand Refractometer mean readings for the plot samples of each seedling were therefore correlated with the corresponding figures for seedling sucroses. It is seen on Table III that the coefficients are high and definitely significant. This, even when small numbers of seedlings only are used.

The additional advantage of the use of the Zeiss Hand Refractometer, particularly in second year seedlings, and by the method of taking three samples from individual canes, is, that it provides information on the stage of juice maturity in seedlings. This applies especially to the early reaped group. This information is naturally unobtainable by the usual sucrose determinations from plot sample bundles. A knowledge of the stage of maturity in seedlings would prove very useful in selecting, and more particularly in allocating, selected seedlings to subsequent trials.

One further aspect of this investigation remains to be discussed.

All correlation work deals with populations of seedlings. Seedling selection work is concerned with comparisons of individuals. In regard to the feature used in selection work—sucrose in juice, it has been shown by correlation in seedling populations that the Zeiss Hand Refractometer brix determinations show remarkably good positive coefficients with sucrose per cent. in juice. In how far, however, can the Zeiss Hand Refractometer indicate the sucrose per cent. in juice of individual seedlings and be of service in selection work?

The target diagrams answer this question. Two such diagrams are illustrated here in Figures II & III. Figure II shows the distribution of the relationship between the Zeiss Hand Refractometer brix and sucrose in juice of the first year seedlings. Assuming that, in this group of seedlings of early proven crosses, the minimum brix reading for selection consideration in seedlings, had been twenty per cent., it is seen that all seedlings in this category, with one exception, were shown to have sucrose percentages of 14.5 upwards. When the sucrose sampling error of single stools of cane is considered, it is concluded that the Zeiss Hand Refractometer Readings in first year seedlings give, for selection purposes, as workable a reading for individual seedlings as the sucrose per cent. in juice.

In actual selection work, naturally, comparison would, as usual, be made between seedlings derived from restricted areas.

Figure III shows the distribution of the relationship between the Zeiss Hand Refractometer brix and sucrose per cent. in juice of thirteen second year seedlings. For purposes of selection, and in so far as juice quality is concerned in selection, it is seen that a very reliable indication of the comparative sucroses of individual seedlings would have been obtained from the Zeiss Hand Refractometer brixes.

V. Conclusions.

The main conclusion from the foregoing investigations is that the use of the Zeiss Hand Refractometer furnishes a comparatively simple, and, by certain methods, a reliable means of indicating seedling sucroses in so far as this feature is used in seedling selection work.

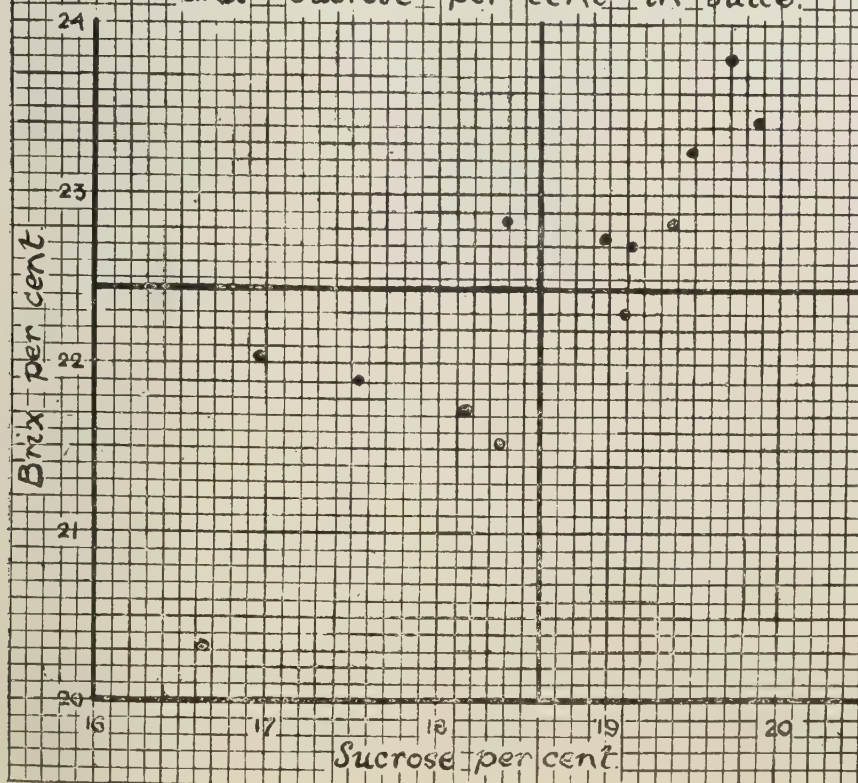
In first year seedlings and in proven crosses (i.e. noble cane seedlings), it appears that three canes sampled from the stool give a sufficiently good indication of the sucrose per cent. It would appear that, in the case of unripe seedlings, it is particularly advisable to take top cane samples, while in over-ripe seedlings it is better to eliminate bottom cane samples.

Thus, the best procedure would appear to be as follows: For early seedlings, select three average sound canes, and make three samples in each, i.e. top, middle and bottom—a total of nine samples. For late and probably over-ripe seedlings, the graph shown in Fig. I would strongly suggest the best procedure would be to select four canes and for each take a top and middle sample—a total of eight samples.

FIG III.

SECOND YEAR SEEDLINGS TODDS.

Target Diagram showing the relationship between Mean Seedling Hand Refractometer Brix and Sucrose per cent in Juice.



In the case of experimental crosses, which are reaped about mid-crop, it would appear to be better to use four canes, and sample each at top, middle and bottom—a total of twelve samples.

In second year seedlings, a more accurate comparison of seedling sucrose is possible: this on account of replications overcoming, to a certain extent, the effect of soil heterogeneity on sucrose development, and at the same time admitting of statistical enquiry.

The procedure which would appear to satisfy best a legitimate comparison of seedling sucroses would consist in taking, for the early group, three average sound canes from each plot and sampling each at three places, i.e., top, middle and bottom—a total of nine samples per plot, and for the late group, four canes of each plot and two samples from each, i.e., middle and top—a total of eight samples. The mean brix reading of the nine and eight samples respectively would serve as the brix reading for the canes in the plot. The seedling means would thus be derived from the mean brix of four plots. The usual statistical analyses and establishment of significant differences between seedling brix means could then be done.

In the first year seedlings, the use of the Zeiss Hand Refractometer would eliminate the necessity of sending half-bundles to the Laboratory, and thereby save planting material in the case of seedlings eventually selected. This is an important advantage in the routine work of propagating selections.

A further advantage of the use of the Zeiss Hand Refractometer in indicating sucroses lies in the fact that, owing to simplicity in usage, the number of seedlings actually tested may be greatly increased without an increase in labour over that used previously, when half bundles were sent to the laboratory for sucrose determinations. It follows that, for purposes of contrasting populations in regard to sucrose inheritance, many more seedlings of each population could be sampled and thus greater accuracy would accrue.

VI. Future Lines of Work.

In so far as the use of the Zeiss Hand Refractometer in indicating the sucroses in first and second year seedlings is concerned, the results of this investigation point to future lines of work being directed towards the practical application of the conclusions noted in the previous section.

The actual work of sampling first year seedlings will in future be delegated to the field officers in charge of reaping gangs. In general, it may be said that these officers will in future reapings of first and second year seedlings employ the time previously used in preparing sample bundles for the laboratory, in obtaining Zeiss Hand Refractometer brix readings.

VII. Summary.

- (1) The investigations described in this paper were undertaken to test the degree of reliability of the Zeiss Hand Refractometer in indicating sucrose percentages in sugar cane first and second year seedlings.

- (2) Twelve samples were taken from each of approximately 200 seedlings, in each of the early and late proven cross groups, and the experimental cross group in the first year seedlings. In the second year seedlings, grown under two environments, twenty-one samples were taken from each sampled plot.
- (3) In the first year seedlings, the stool was the unit. Three samples were taken in each of four canes, the sample being taken from the middles of internodes, approximately one foot from the top and the bottom and at the middle of the cane.

In the second year seedlings, the plot was the unit, and the routine laboratory plot sample bundle was used to provide seven canes selected at random. Three samples were taken from each of the seven canes.

- (4) The comparative efficiency of various groupings of samples was ascertained by correlation with appropriate sucrose in juice percentages. These correlations are presented in Section III.
- (5) The effects of the factors (1) number of canes sampled, (2) position of sample on cane, (3) time of reaping and (4) nature of the seedlings sampled, on the strength of the correlation between Zeiss Hand Refractometer brix and sucrose in juice are discussed.
- (6) All correlations established are seen to be high, although certain sampling combinations are better than others.
- (7) By reference to target diagrams, it is seen that the use of the Zeiss Hand Refractometer is comparatively safe in indicating the sucrose of individual seedlings.
- (8) The main conclusion is that, by certain sampling methods, the Zeiss Hand Refractometer may safely be used in indicating sucroses in seedlings, and substituted for actual sucrose determinations in seedling selection work. The considered best methods in first year proven crosses and experimental crosses, and in second year seedling trials, are noted.
- (9) Future lines of work will be directed towards the practical application of the findings of this investigation in first and second year sugar cane seedling trials.

VIII. References to Literature.

- (1) Craig, N. (1931) The Use of the Refractometer in Cane Seedling Selection Work. *Int. Sugar Jour.*, XXXIII, p. 14.
- (2) do. (1934) Use of the Zeiss Sugar Hand Refractometer in Sugar Cane Selection Work. *Ibid* XXXVI. p. 235.
- (3) Levert, L., (1932) *Arch. Suikerind. Ned.—Indie XL*, 26.
van der Wonde, C. A. A.
&
van Dillewijn, C. (See abstract in *Facts about Sugar*, Vol. XXVII, No. 10).
- (4) McIntosh, A. E. S. (1935) An Account of the Policy and Methods Employed in the Breeding and Initial Stage of Selecting Sugar Cane Seedlings. B.W.I. Central Sugar Cane Breeding Station, Bull. 6.
- (5) do. (1935) An Account of the Methods Employed in Testing and Selecting Sugar Cane Seedlings for Thick-Cane Areas in the British West Indies. *Ibid*, Bull. 9.
- (6) do. (1933) *Agric. Journ., Dept. Sci. & Agric., Barbados*, Vol. 2, No. 3.
- (7) Verret, J. A. (1928) Use of the Hand Refractometer in the Selection of Seedlings. *Hawaiian Planters' Record* XXXII, p. 427.
&
Mangelsdorf, A. J.

